



University of Padova, Italy
Interdepartmental Research
Center of Myology (CIR Myo)



Physiko&Rheumatherapie, Inst. f. Phys. Med.,
St. Pölten &
Ambulante Reha, Amstetten, Austria

Easy Aging



A&C M-C Foundation
for Translational
Myology

2020 PaduaMuscleDays, November 25-28, Euganei Hills and Padova, Italy

MOBILITY MEDICINE: 30 YEARS OF TRANSLATIONAL RESEARCH

Euganei Hills, Padova (Italy), November 19 - 21, 2020

Hotel Petrarca, Piazza Roma 23, Montegrotto Terme, Euganei Hills (Padova) 35122 Italy

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Organizers: Ugo Carraro, Helmut Kern, Christiaan Leeuwenburgh, Marco Narici, Feliciano Protasi, Marco Sandri, Lee Sweeney



THURSDAY November 19, 2020

2020 Padua Muscle Days go Virtual: Italy Morning = Japan Afternoon

Italy 09.20 AM; Iceland 08.20AM; USA ET 02.20 AM; Japan 04.20 PM

Italy 09:20 AM **2020 Padua Muscle Days during the COVID-2020 Outbrek**, Ugo Carraro

Japan 04:30 PM Challenge of Osato Research Institute for Preventive Medicine with the aim to reduce medical costs in super-aging societies, Yuki Hayashi, Gifu, Japan

Japan 05:00 PM Dietary Phosphorus Overload Aggravates the Phenotype of the Dystrophin-Deficient mdx Mouse. Therapeutic effect of calcium and Vitamin C on muscle degeneration in elevated Pi or at low temperature cultures, Ryoichi Matsuda, Department of Life Sciences, University of Tokyo, Japan

Italy 10:30 AM The DLK1-DIO3 cluster miRNAs regulate mitochondrial functions in the dystrophic muscle in DMD, Ai Vu Hong, Nathalie Bourg, Peggy Sanatine, Jerome Poupiot, Karine Charton, Evelyne Gicquel, Marco Spinazzi, Isabelle Richard, David Israeli, Evry, France

Italy 11:00 AM Single-cell analysis revealed the importance of non-coding RNAs for muscle plasticity, Stefano Cagnin, Biology, University of Padova, Italy

Italy 11:30 AM Relationship between ETFDH mutations, expression levels of serum myomiRNAs and response to treatment in MADD patients, Sara Missaglia, et al., Milan, Padova, Venice, Italy

Italy 12:00 AM Blood contamination of human mouth fluid: a non-invasive approach for serological analyses in old and very old persons, Barbara Ravara, et al., A&C M-C Foundation, Padova, Italy



THURSDAY November 19, 2020

2020 Padua Muscle Days go Virtual: Italy Afternoon = USA Morning

Italy 03.00 PM; Iceland 02.00 PM; USA ET 08.00 AM; Japan 10.00 PM

USA ET 08:00 AM

SARCOPENIA AND AGING: Nutritional, Pharmacological and Physiological Interventions

Christiaan Leeuwenburgh, Russ Hepple, Chairs

USA ET 8:00 AM

Effects of epicatechins on endothelial, mitochondrial and physical function: Clinical Trial Results. *Christiaan Leeuwenburgh, University of Florida, Gainesville, FL, USA*

USA ET 8:30 AM

The impact of Fermented Papaya Product (FPP) on cognitive and brain function in older adults: a pilot clinical trial, *Adam J. Woods, University of Florida, Gainesville, FL, USA*

USA ET 9:00 AM

Mitochondrial Permeability Transition causes muscle atrophy in advanced age, *Russel T. Hepple, University of Florida, Gainesville, USA*

USA ET 9:30 AM

Neuromuscular activity of *C. elegans* and zebrafish models of *FBXL4*-based mitochondrial respiratory chain disease: translational platforms for drug screening, *Manuela Lavorato et al., Department of Pediatrics, The Children's Hospital of Philadelphia, Philadelphia, PA, USA*

Italy 5:00 PM

Impact of ageing and exercise on the neuromuscular junction, *Marco Narici, Department of Biomedical Sciences, Padua University, Italy*

Italy 5:30 PM

Neurohypophyseal hormones and skeletal muscle: a tale of two faces, *Sergio Adamo, Sapienza University of Rome, Italy*

Italy 6:00 PM

A role for fermented papaya in preventing and treating aging and cancer cachexia, through a potent and systemic antioxidant effect, *Mariantonia Logozzi et al., Istituto Superiore di Sanità, Rome, Italy*

USA ET 11:30 AM

Prospects for exercise training in the absence of muscle IGF-1, *Elisabeth R. Barton, Dept. of Applied Physiology & Kinesiology, University of Florida, Gainesville, USA*

Italy 7:00 PM

A possible strategy to prevent skeletal muscle atrophy induced by immobilization, *Paula Tavares, Faculty of Sport Sciences and Physical Education, University of Coimbra, Portugal*



Hotel Petrarca, Euganei Hills, (Padova), Italy

FRIDAY November 20, 2020

Virtual-Presentations: Italy Morning = Japan Afternoon

Italy 09.00 AM; Iceland 08.00 AM; USA ET 03.00 AM; Japan 14.00 PM

- Italy 9:30 AM ***Commitment to reproducibility in targeting mitochondrial respiratory control: basic and advanced applications of the O₂k-FluoRespirometer***
- Italy 9:20 AM Forewords, *Ugo Carraro, A&C M-C Foundation, Padova, Italy*
- Italy 9:30 AM Commitment to reproducibility in mitochondrial respiration studies with permeabilized muscle fibers, *Carolina Doerrier, Marco Di Marcello, Erich Gnaiger, Oroboros Instruments, Innsbruck, Austria*
- Italy 10:30 AM Mitochondrial ROS production, *Nina Kaludercic, CNR Neuroscience Institute, University of Padova, Italy*
- Italy 11:00 AM Age-related changes of mechanical stiffness of extra-cellular matrix in human skeletal muscle, *Lorenzo Marcucci, et al., Department of Biomedical Sciences, Padua University, Italy*
- Italy 11:20 AM Variability and inter rater reliability of ultrasound imaging of fasciae/muscles, *Carmelo Pirri, et al., Dept. Neurosciences, University of Padova, Italy*
- Italy 11:40 AM Advances in imaging techniques for the study of human skeletal muscle in-vivo, *Martino Franchi, Department of Biomedical Sciences, Padua University, Italy*
- Italy 12:00 AM Testosterone therapy in Alzheimer's disease, *Vittorio Emanuel Bianchi, Department of Endocrinology and Metabolism, Falciano, San Marino*



Hotel Petrarca, Euganei Hills, (Padova), Italy

FRIDAY November 20, 2020

Virtual-Presentations: Italy Afternoon = USA Morning

Italy 1:40 PM; Iceland 0:40 PM; USA ET 7:40 AM; Japan 7:40 PM

- Italy 1:40 PM **Translational Mobility Medicine Lecture 1.**
The genetic underpinning of V_{O2max} and trainability, *Hans Hoppeler, Anatomy Institute, University of Berna, Switzerland*
- Italy 2:20 PM **Translational Mobility Medicine Lecture 2.**
Ear Stimulation, from Padua 1600 to cochlear implants, *Alessandro Martini, Neuroscience Department, Padua University, Italy*
- Italy 3:00 PM **News on EEG, EMS, FES, TMS and more,**
Helmut Kern, Alessandro Martini, Chairs
- USA ET 9:00 AM Remediating age-related cognitive and physical decline with transcranial direct current stimulation (tDCS), *Adam J. Woods, University of Florida, Gainesville, USA*
- Italy 3:20 PM Characterisation of diabetic myopathy by high density EMG, *Giuseppe De Vito, CIR-Myo & Dept. Biomedical Sciences, Padua University, Italy*
- Italy 3:40 PM Body mass excess, muscle mass, obesity and mitochondrial fitness, *Erich Gnaiger, Medical University of Innsbruck and Oroboros Instruments, Innsbruck, Austria*
- Iceland 3:00 PM Using high density EEG to assess TMS treatment in patients with schizophrenia, *Ovidiu C Banea et al., Institute for Biomedical and Neural Engineering, Reykjavík University, Reykjavík, Iceland and Landspítali, Iceland*



Hotel Petrarca, Euganei Hills, (Padova), Italy

FRIDAY November 20, 2020

Italy 4:20 PM; Iceland 3:20 PM; USA ET 12.00 AM; Japan 11.20 PM

Italy 4:20 PM **Therapies for genetic diseases,**
Sweeney L, Tavian D, Chairs

USA ET 10.20 AM Gene Therapies for Duchenne Muscular Dystrophy, *Lee Sweeney, Myology Institute, University of Florida, Gainesville, USA*

Italy 4:40 PM PABPN1 nuclear aggregates in oculopharyngeal muscular dystrophy, *Gillian Butler-Browne, Sorbonne Université, Paris, France*

Italy 5:00 PM Mitochondria as targets in Duchenne Muscular Dystrophy, *Paolo Bernardi, Department of Biomedical Sciences, University of Padova, Italy*

Italy 5:20 PM **Mobility Medicine Imaging,** *Feliciano Protasi, Ugo Carraro, Chairs*

Italy 5:20 PM Discovery of Calcium Entry Units: when electron microscopy still counts, *Feliciano Protasi, Chieti University, Italy*

USA ET 11.40 AM HERG Expression in C2C12 Myotubes leads to upregulation of genes related to Interferon gamma, *Amber Pond, Southern Illinois University School of Medicine, USA*

Italy 6:00 PM Formation of Tubular Aggregates in muscle: role of STIM1 and Orai1, *Simona Boncompagni, Chieti University, Italy*

Italy 6:20 PM **Mobility Medicine, Clinical imaging,** *Marco Narici, Paolo Gargiulo, Chairs*

USA WT 7.20 AM Skeletal muscle mechanics in the aging muscle: Advanced Fast MRI technologies provide insights into skeletal muscle dynamics and physiology, *Shantanu Sinha, University of California at San Diego, CA, USA*

USA WT 7.40 AM Bye, Bye Biopsy: Extracting muscle tissue composition and microstructure from Magnetic Resonance Imaging (MRI), initial validation to biopsy, and application to the aging muscle, *Usha Sinha, San Diego State University, San Diego, CA, USA*

Italy 7:00 PM Biomarkers of muscle atrophy and of neuromuscular maladaptation during 10-day bed rest. *Marco Narici, CIR-Myo, Department of Biomedical Sciences, Padua University, Italy*

Iceland 6:20 PM Predicting cardiovascular pathophysiology from a mid-thigh CT image, *Paolo Gargiulo, et al., Biomedical and Neural Engineering, Reykjavík University, Iceland*

USA ET 3:40 PM Soft tissue radiodensity, self-reported physical activity, and lower extremity function in the AGES-Reykjavík study, *Kyle Edmunds et al., Reykjavík University, Iceland*



Hotel Petrarca, Euganei Hills, (Padova), Italy

SATURDAY November 21, 2020

Italy 9:00 **The Center of Active Aging** - *Helmut Kern, Chair*

- 9:00 AM Research in rehabilitation, past & future programs, *Helmut Kern, Wien, Austria*
- 9:20 AM Centre of Active Ageing: Current status, *Stefan Loeffler, Wien, Austria.*
- 9:40 AM 15 years of Vienna-Padova-Chieti collaboration: what did we learn, *Feliciano Protasi, University of Chieti, Italy*
- 10:00 AM Signals from the niche to modulate muscle regeneration, *Antonio Musarò, Sapienza University of Rome, Italy*
- 10:20 AM Resistance training as supplement therapy in hypogonadal men, *Milan Sedliak, Comenius University, Bratislava, Slovakia*
- 10:40 AM Evaluation of sympathetic arousal by skin conductance measurement: A tool to optimize rehabilitation strategies? *Manfred Bijak, Center for Medical Physics and Biomedical Engineering, MedUni Vienna, Austria*
- 11:00 AM **The Center of Active Aging – Brainstorming on the Future**
Hans Oppeler, Lars Larsson, Chairs
Discussants: *Mauro Alaibac, Giovanna Albertin, Simona Boncompagni, Ugo Carraro, Jan Cvecka, Dusan Hamar, Cristian Hofer, Helmut Kern, Stefan Loeffler, Antonio Musarò, Feliciano Protasi, Barbara Ravara, Nejc Sarabon, Sascha Sajer, Milan Sedliak, Veronika Tirpakova, Sandra Zampieri, Attendees ...*
- 12:30 AM **Translational Mobility Medicine Lecture 3.**
Neurogenic vs. myogenic origin of acquired muscle paralysis in ICU patients: Evaluation of different diagnostic methods
Lars Larsson, Karolinska Institutet, Stockholm, Sweden



Hotel Petrarca, Euganei Hills, (Padova), Italy

SATURDAY November 21, 2020

- 2:00 PM **Mobility Disorders & Rehabilitation I**, D Coletti, G Fanò, Chairs
- 2:00 PM Treatment of Central Core Disease with Functional Electrical Stimulation (FES): a Case Report, *Feliciano Protasi, University of Chieti, Italy*
- 2:20 PM Modulation of some vital functions in a patient with angina pectoris using transcutaneous auricular nerve stimulation, *Janez Rozman, et al, University of Ljubljana, Slovenia*
- 2:40 PM Medical Emergency in critical environment: Physical capacities of Emergency Team, *Francesco Coscia, Paola V. Gigliotti, Rezhna Adil Rasheed, Giorgio Fanò-Illic, Perugia, Italy*
- 3:00 PM Sensitivity of the fasciae to endocannabinoid system and remodeling of fascial matrix: consequences for fascial fibrosis and inflammation, *Caterina Fede et al., Dept. Neurosciences, University of Padova, Italy*
- 3:20 PM Comparison of reflex period in pendulum test done in SCI and Stroke patients, *Thordur Helgason et al., Reykjavík University, Iceland*
- 3:40 PM Skin and mouth fluids analyses to evaluate biological age in older and oldest persons, *Giovanna Albertin et al., Dept. Neuroscience, University of Padova, Italy*
- 4:30 PM **Mobility Disorders & Rehabilitation II** C Angelini, S Masiero, Chairs
- 4:30 PM Central Myonuclei and denervation markers in Cancer Cachexia, *Dario Coletti, Sapienza University of Rome, Italy*
- 5:00 PM Comparison of morphological and serological analyses of denervation biomarkers in skeletal muscle wasting conditions, *Sandra Zampieri, DiSCOG, University of Padova, Italy and Austria*
- 5:00 PM Muscle activity prevents the uncoupling of mitochondria from Ca²⁺ release units induced by ageing and disuse, *Laura Pietrangelo et al., Chieti University, Italy*
- 5:20 PM Circulating microRNAs as promising biomarkers for monitoring of NLSDM clinical phenotype, *Valentina Pegoraro, Roberta Marozzo, Sara Missaglia, Daniela Tavian, Corrado Angelini, Padova Venice, Milan, Italy*
- 5:40 PM Exercise-activated Ca²⁺ entry and enhanced risk of Heat Stroke. *Barbara Girolami, et al. University G. d'Annunzio of Chieti-Pescara, Chieti, Italy*
- 6:00 PM Methods to monitor mitochondrial activity in skeletal muscle, *Gaia Gheradi, et al., Department of Biomedical Sciences, University of Padova, Italy*
- 6:20 PM **Zipora Yablonka-Reuveni, Ugo Carraro: See you to 2021 (Virtual) Padua Muscle & Mobility Medicine Days (2021 V-PM3Ds), March 25-27 - Euganei Hills (Padova), Italy**

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unit (ICU) patients. ICU patients with acquired quadriplegia in response to critical care were included in the study. A total of 142 patients were examined with routine electrophysiological methods, together with biochemical analyses of myosin actin (M:A) ratios of muscle biopsies. In addition, the comparison of evoked EMG response in direct vs. indirect muscle stimulation and histopathological analyses of muscle biopsies were performed in a subset of the patients. ICU patients with quadriplegia were stratified into five groups based on the hallmark of CIM, i.e., preferential myosin loss (myosin:actin ratio, M:A) and classified as severe (M:A < 0.5; n= 12), moderate (0.5 ≤ M:A < 1; n= 40), mildly moderate (1 ≤ M:A < 1.5; n=49), mild (1.5 ≤ M:A < 1.7; n= 24) and normal (1.7 ≤ M:A; n=19). Identical M:A ratios were obtained in the small (4-15 mg) muscle samples using a disposable semiautomatic micro biopsy needle instrument as in the larger (>80 mg) samples obtained with a conchotome instrument. Compound muscle action potential (CMAP) duration was increased and amplitude decreased in patients with preferential myosin loss but deviations from this relationship were observed in numerous patients resulting in only weak correlations between CMAP properties and M:A. Advanced electrophysiological methods measuring refractoriness and comparing CMAP amplitude after indirect nerve vs. direct muscle stimulation are time consuming and did not increase precision compared with conventional electrophysiological measurements in the diagnosis of CIM. Low CMAP amplitude upon indirect vs direct stimulation strongly suggests a neurogenic lesion, i.e., CIP, but this was rarely observed among the patients in this study. Histopathological diagnosis of CIM/CIP based on enzyme-histochemical mATPase stainings were hampered by poor quantitative precision of myosin loss and the impact of pathological findings unrelated to the acute quadriplegia. Conventional electrophysiological methods are valuable in identifying a peripheral origin of quadriplegia in ICU patients, but do not reliably separate between neurogenic vs. myogenic origin of paralysis. The hallmark of CIM, the preferential myosin loss, can be reliably evaluated in the small samples obtained with the micro biopsy instrument. The major advantage of this method is that it is less invasive than conventional muscle biopsies, reducing the risk of bleeding in ICU patients frequently on anticoagulant treatment, and it can be repeated multiple times during follow up for monitoring purposes.

Keywords: Critical care, myosin, myopathy, ENeG, EMG, CMAP, muscle biopsy

References

1. Friedrich O, Reid MB, Van den Berghe G, et al., *The Sick and the Weak: Neuropathies/Myopathies in the Critically Ill*. *Physiol Rev* 2015;95:1025-109. doi: 10.1152/physrev.00028.2014
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3. Larsson L, Li X, Edström L, et al., *Acute quadriplegia and loss of muscle myosin in patients treated with nondepolarizing neuromuscular blocking agents and corticosteroids: mechanisms at the cellular and molecular levels*. *Crit Care Med* 2000;28:34-45.
4. Stibler H, Edström L, Ahlbeck K, et al. *Electrophoretic determination of the myosin/actin ratio in the diagnosis of critical illness myopathy*. *Intensive Care Med* 2003;29: 1515-27. doi: 10.1007/s00134-003-1894-9
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6. November 21, 2020 – Italy Time 3:00 PM

Central Myonuclei and denervation markers in Cancer Cachexia

Nissrine Daou (1), Medhi Hassani (1,2,3), Emidio Matos (4), Gabriela Salim De Castro (5), Raquel Galvao Figueredo Costa (5), Marilia Seelaender (5), Viviana Moresi (2,3), Marco Rocchi (6), Sergio Adamo (2, 3), Zhenlin Li (1), Onnik Agbulut (1), Dario Coletti (1,2,3)*

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An idiopathic myopathy characterized by central nuclei in muscle fibers, a hallmark of muscle regeneration, has been observed in cancer patients. In cancer cachexia skeletal muscle is incapable of regeneration, consequently, this observation remains unaccounted for. In C26-tumor bearing, cachectic mice, we observed muscle fibers with central nuclei in the absence of molecular markers of bona fide regeneration. These clustered, non-peripheral nuclei were present in NCAM-expressing muscle fibers. Since NCAM expression is upregulated in denervated myofibers, we searched for additional markers of denervation, including AchRs, MUSK, and HDAC. This last one being also consistently upregulated in cachectic muscles, correlated with an increase of central myonuclei. This held true in the musculature of patients suffering from gastrointestinal cancer, where a progressive increase in the number of

central myonuclei was observed in weight stable and in cachectic patients, compared to healthy subjects. Based on all of the above, the presence of central myonuclei in cancer patients and animal models of cachexia is consistent with motor neuron loss or neuromuscular junction perturbation and could underlie a previously neglected phenomenon of denervation, rather than representing myofiber damage and regeneration in cachexia. Similarly to aging, denervation-dependent myofiber atrophy could contribute to muscle wasting in cancer cachexia.

Keywords: Central Myonuclei, denervation markers, cancer cachexia

References

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7. November 21, 2020 – Italy Time 6:40 PM

Exercise-activated Ca²⁺ entry and enhanced risk of Heat Stroke

Barbara Girolami, Laura Pietrangelo, Antonio Michelucci, Matteo Serano, Feliciano Protasi*

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Exertional/Environmental Heat Strokes (EHSs) are hyperthermic crises triggered by strenuous physical exercise and/or exposure to environmental heat, which are caused by an altered intracellular Ca²⁺ homeostasis in muscle (Bouchama and Knochel, 2002).¹ Store-Operated Ca²⁺ Entry (SOCE) is a mechanism that influences intracellular Ca²⁺ levels, allowing recovery of

extracellular Ca²⁺ during prolonged activity. We recently demonstrated that exercise leads to formation of Calcium Entry Units (CEUs), intracellular junctions between stacks of sarcoplasmic reticulum (SR) and transverse tubules (TTs) at the I band that promote interaction between STIM1 and Orai1, the two proteins that mediate SOCE (Boncompagni et al. 2017; Protasi et al. 2020).^{2,3} Here we tested the hypothesis that exercise-induced assembly of CEUs may increase the risk of hyperthermic crisis when physical activity is performed in challenging environmental conditions. 4 months old mice were: a) first, divided in 3 experimental groups: control, trained-1m (1 month of voluntary running in wheel cages), and exercised-1h (1 hour of incremental treadmill run); and b) second, subjected to an incremental treadmill run of 45 min at 34°C and 40% humidity. We then: a) measured the internal temperature of mice, which was higher in the pre-exercised groups (trained-1m: 38.9°C ± 0.33; exercised-1h: 38.7°C ± 0.40) compared to control (37.9°C ± 0.17). b) applied an ex-vivo exertional stress protocol to isolated EDL muscles (tetanic stimulation performed at 30°C) and verified that samples from trained-1m and exercised-1h mice generated a tension significantly greater than control. c) Analyzed CEUs by electron microscopy (EM) and verified that EDL muscles of exercised-1h and trained-1m mice contained a greater number of elements forming CEUs. The data collected suggest that assembly of Calcium Entry Units during exercise could predispose to EHS when exercise is performed in challenging environmental conditions.

Keywords: Exercise-activated Ca²⁺ entry, risk of heat stroke, challenging environmental conditions

References

1. Bouchama A, Knochel JP. Heat stroke. *N Engl J Med* 2002;346:1978. doi: 10.1056/NEJMra011089
2. Boncompagni S, Michelucci A, Pietrangelo L, et al. Exercise-dependent formation of new junctions that promote STIM1-Orai1 assembly in skeletal muscle. *Sci Rep* 2017;7(1):14286. doi: 10.1038/s41598-017-14134-0
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List of acronyms

AD - Alzheimer's disease
 CEUs - Calcium Entry Units
 CIM - Critical Illness Myopathy